SimpliNano™

Product User Manual

Codes: 29061711 SimpliNano
29061712 SimpliNano with printer
29061713 SimpliNano (North American version)
29061714 SimpliNano with printer (North American version)

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1. INSTALLATION

1.1. Unpacking and positioning

- It is recommended that users read through this manual prior to use.
- Remove the instrument from its packaging and inspect it for signs of damage. If any are discovered, inform your supplier immediately.
- The instrument must be placed on a stable, level surface that can take its weight (~ 4.5 kg) and positioned such that air can circulate freely around the casing.
- Ensure your proposed installation site conforms to the environmental conditions for safe operation.
- The instrument is designed for indoor use only, temperature range 5°C to 35°C and should be kept away from strong draughts.
- If you use the instrument in a room subjected to extremes of temperature change during the day, it may be necessary to recalibrate (by switching off and then on again) once thermal equilibrium has been established (2–3 hours).
- A temperature change of no more than 4°C/hour and a maximum relative humidity of 80% at 31°C, decreasing linearly to 50% at 40°C are required.
- If the instrument has just been unpacked or has been stored in a cold environment, it should be allowed to come to thermal equilibrium for 2–3 hours in the laboratory before switching on. This will prevent calibration failure as a result of internal condensation.
- The instrument must be connected to the power supply with the power adaptor supplied. The adaptor can be used on 90–240 V, 50–60 Hz supplies. It will become warm once plugged into the power supply and should not be covered up.
- Switch on the instrument via the keypad (⏻) after it has been plugged in. The instrument will perform a series of self-diagnostic checks.
- Contact your supplier if you experience any difficulties with this instrument.

1.2. Safety

Spectrophotometer Health & Safety Document including General Operating Instructions are available as a booklet provided with each instrument. The booklet is translated into the European languages and is available on the User Manual CD supplied with each instrument. The instructions cover basic operation, troubleshooting and how to use the instrument in a safe manner.

CAUTION

The instruments contain a UV source which generates a light beam that traverses the sample chamber and is accessible in the lamp chamber. Under normal use the lamp beam is confined within the instrument. The unit should not be operated with the lamp housing lid removed as prolonged exposure to the beam may cause permanent eye damage.

WARNING

High voltages exist inside the SimpliNano instruments. Repair and maintenance should only be carried out by individuals trained specifically to work on these instruments.

If the instrument is used in a manner not specified or in environmental conditions not appropriate for safe operation, the protection provided may be impaired and instrument warranty withdrawn.

There are no user-serviceable parts inside this instrument.

1.3. Declaration of conformity

GE certifies that SimpliNano UV/Visible Spectrophotometers; part numbers 29061711, 29061712 conform to the requirements of the following Directives :-

2006/95/EC	Low voltage directive (LVD)
2004/108/EC	Electromagnetic Compatibility (EMC) directive
2012/19/EU	Waste Electrical and Electronic Equipment directive recast (WEEE Recast)
2011/65/EU	Restriction on the use of certain hazardous substances (ROHS) directive
2006/42/EC	Machinery directive

Standards to which conformity is declared are as follows:

EN61010 - 1:2010	Safety requirements for electrical equipment for measurement, control and laboratory use. General requirements
EN61326-1:2006	Electrical equipment for measurement, control and laboratory use - EMC Requirements
EN50581:2012	Technical documentation for the assessment of electrical and electronic products with respect to the restriction of hazardous substances
EN ISO 12100:2010	Safety of machinery - General principles for design, risk assessment and risk reduction

2. INTRODUCTION

2.1. Your Spectrophotometer

SimpliNano is a simple-to-use UV/Visible instrument with twin CCD array detectors (1024 pixels). It has no moving parts, which is the basis of the rapid scanning operating system.

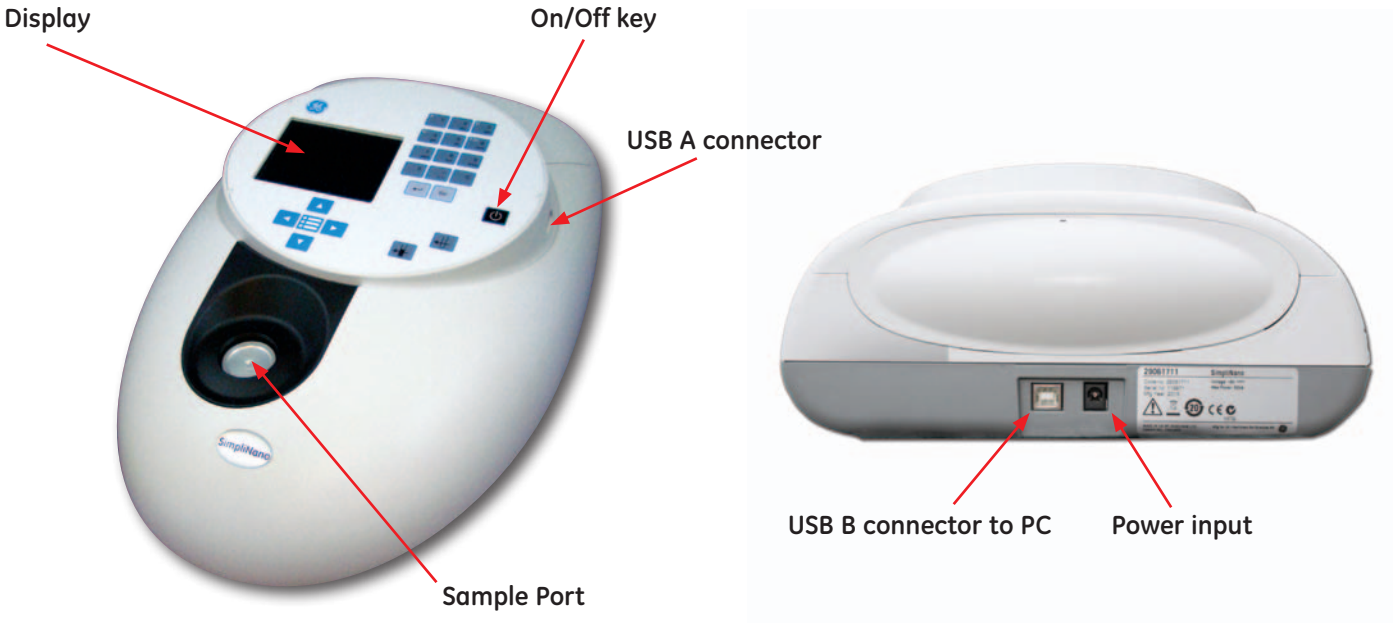


Figure 1: SimpliNano Spectrophotometer

The SimpliNano is fitted with a micro volume sampling port with a fixed 0.5 mm pathlength for measuring low volumes of highly absorbing life science samples such as DNA, RNA and proteins.

2.2. File system

After switch on and automatic checking the instrument defaults to a home page entitled 'SimpliNano'. This page displays seven folders which form the topmost layer of a simple file tree which is the basis of the user-interface. The folder screens are reached by pressing the appropriate number on the keypad, with return to the top level by means of the '***Esc***' key. The folders group various facilities together as follows:-

1. DNA:	Concentration and purity check for DNA samples
2. RNA:	Concentration and purity check for RNA samples
3. Oligo:	Concentration and purity check for Oligonucleotide samples
4. Absorbance Concentration:	Concentration measurement at a single wavelength based on a factor entered or calculated from a single standard.
5. Protein A280:	Concentration and purity check for protein samples
6. Methods:	Saved methods
7. Utilities:	Instrument set up options

2.3. Data export

An integral printer is available for the instrument; this may be either supplied pre-installed or as an optional accessory. The installation procedure is described in section 7.1.

Figure 2: SimpliNano with Printer

Data can be transferred to a PC via a USB™ cable using the supplied software "PVC" (Print Via Computer).

PVC can store data either in a common directory or can be configured to save to independent directories by both file format and connection. The data may be stored as an Excel spreadsheet, an EMF graphics file, a comma delimited (csv) data file, a tab delimited (txt) data file or in native PVC format.

Some users may find it convenient to use PVC to "print through" the PC directly to a local or networked printer.

Datrys Computer control software is also available for SimpliNano as an optional extra.

The SimpliNano USB has the option to save data onto a USB stick in 2 different formats, both formats save Method details, all measured results and all scan data (if available in the application).

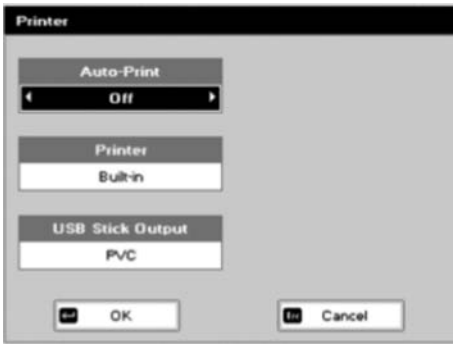
1. PVC format that can only be opened by Print via Computer software from GE Healthcare. Once a file has been opened in PVC it can be exported into ASCII (comma or tab separated), text, Extensible meta format or Excel format (only if Excel is installed on the PC).
2. Tab Separated Variable (TSV) allowing the data to be opened directly into Excel.

Please note that the USB stick needs to be placed into machine BEFORE measurements are made and encrypted sticks & external hard drives are not compatabile with this instrument.

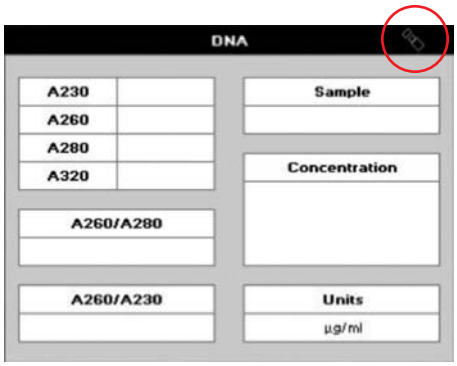
- Insert USB memory stick into the connector on the right hand side of the instrument. An audible click will be heard as the stick is recognised.
- On main menu screen choose ***Utilities*** option by pressing ***7*** on the keypad.
- Then choose ***Printer*** option, by pressing ***3*** on the keypad, to choose ***Data Export Options***.

  Data output can then be made from the ***Printer*** screen. Use arrow keys to scroll down and choose settings.

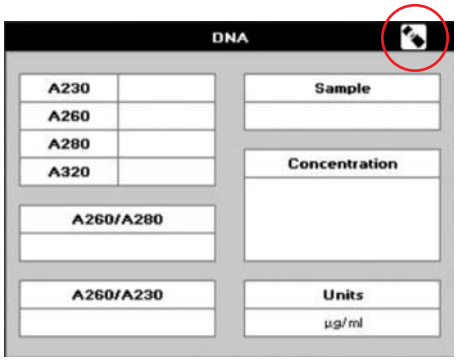
- When the ***Auto-Print*** option is set to OFF all data will automatically be saved to the USB memory stick. The operator can choose either the TSV or PVC format by using the arrows keys to select ***USB Stick Output*** – PVC files can be opened in the PVC program. TSV files can be opened in Microsoft Excel.



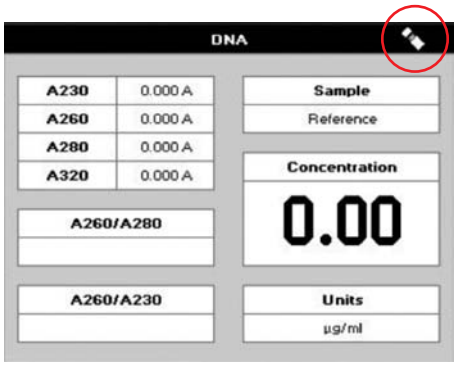
- When a USB stick is inserted and an application selected an icon will appear on the top right hand side of the status bar.



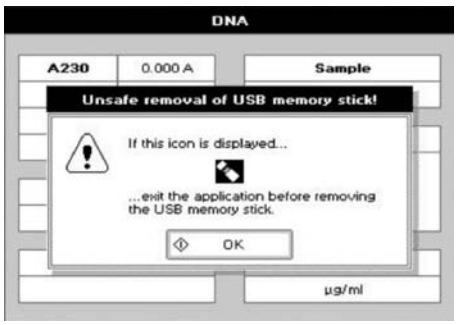
- When the USB stick is being written to the icon will change as shown.



Multiple samples are stored in the same file on the USB stick – during a sample batch the icon will be displayed as shown, to indicate that a file on the USB stick is open but not currently being written to.



The USB stick **should not** be removed when this icon is displayed. To safely remove the USB memory stick after a reading, close the application by first pressing the ***ESC*** key and withdraw the stick from the port WARNING DATA MAY BE LOST IF THE USB STICK IS REMOVED INCORRECTLY. This will be indicated by the following notification.



Data transfer to USB can be carried out simultaneously with printing to a built in printer *(if fitted)* or export to a PC running PVC software *(if connected)*.

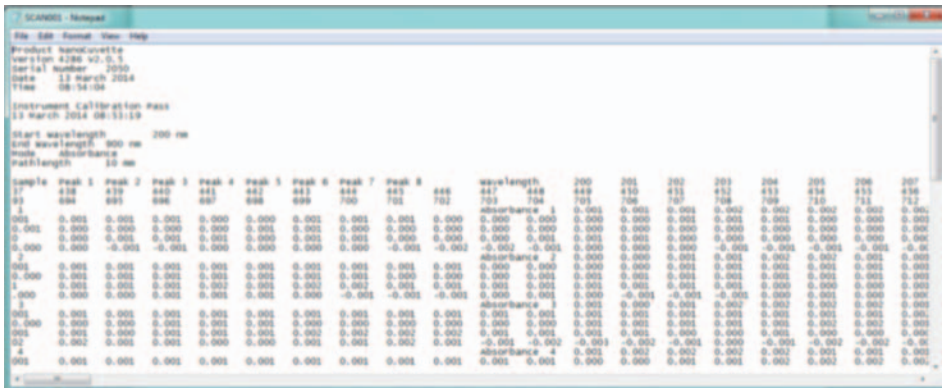
- To transfer data simultaneously to PC, enter ***Printer*** options screen, use arrow buttons on keypad to scroll th change settings as follows:
  - Auto-Print setting: **ON**
  - Printer: **Computer (USB)**
  - USB Stick Output: **TSV or PVC**
- To transfer data simultaneously to Printer, enter ***Printer*** options screen, use arrow buttons on keypad to scroll th change settings as follows:
  - Auto-Print setting: **ON**
  - Printer: **Built-in**
  - USB Stick Output: **TSV or PVC**

2.4. Working with data stored on a USB stick

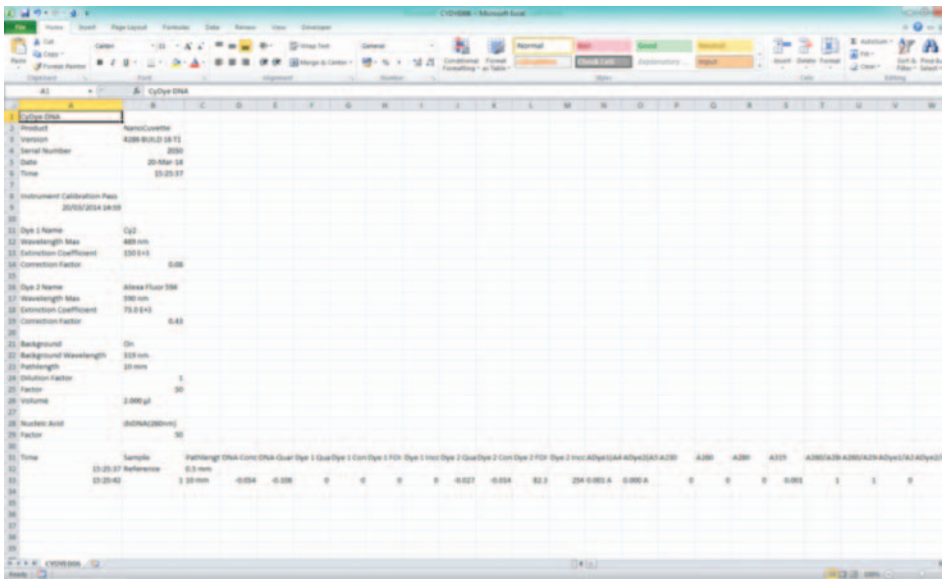
For data stored in PVC fromat please refer to the PVC user manual or help file within PVC software.

For data stored in TSV format insert the USB stick into a PC and use Windows Explorer to navigate to the saved file which will be located in a folder named as the instrument serial number. The file can then be opened in several ways:

1. Double clicking on the .txt file will open the file into Windows Notepad as follows.



2. Select the file, right mouse click and select ***Open With*** from the Menu and select ***Microsoft Excel***. Data will then open directly into Excel.



3. SAMPLE HANDLING

SimpliNano uses a simple sample port which enables users to quickly and accurately measure samples without any moving parts. The sample port comprises of 2 fibre optics which are fixed 0.5 mm apart. The sample should be loaded into this space.

Step 1
Before measurement, inspect the sample port to ensure it is clean.

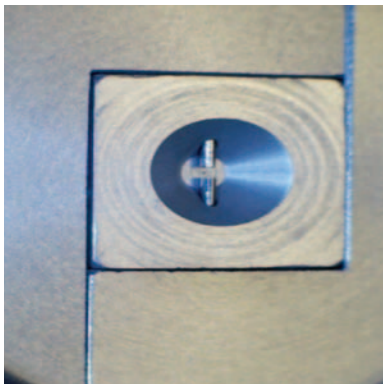


Figure 3a

Figure 3b

Figure 3a and 3b: Sample port area

Step 2
Pipette the reference solution into the port. A good reference will use the same solvent as the sample. To ensure the correct placement of the solution in the port, touch the pipette tip to the bottom of the sample reservoir and then slowly dispense the solution.

Step 3
The sample or reference should be pipetted into the centre of the port and be in contact with both port probes. Take care not to introduce bubbles into the sample.

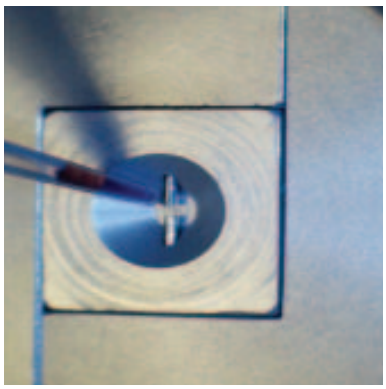


Figure 4a

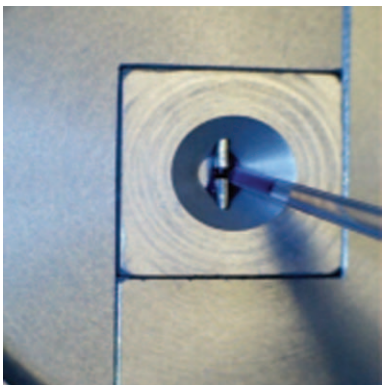


Figure 4b

Figure 4a and 4b: How to correctly load sample onto sample port

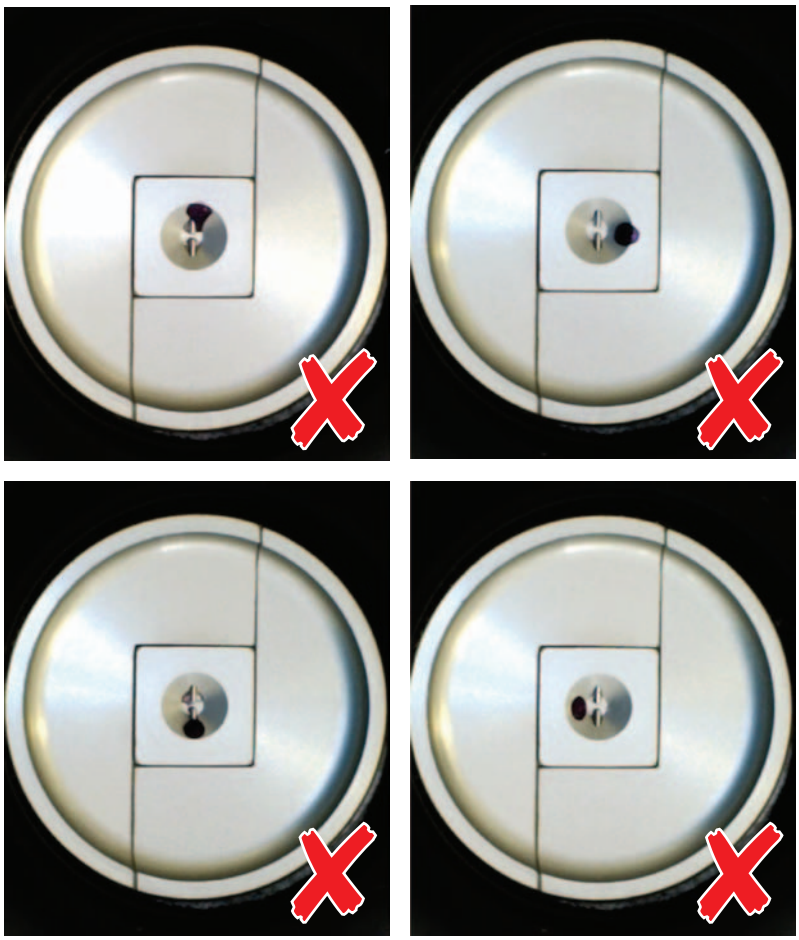


Figure 5

Figure 5: The images display the incorrect loading position of samples

Figure 6

Figure 6: The position and shape of the drop shown in the image is the optimum position for the sample measurement to be made.

Step 4

To clean the the port, simply wipe with tissue or a lint free cloth. When cleaning the port, it is ok to touch the probes.

Once the port is clean, you are ready to proceed with your next sample measurement.

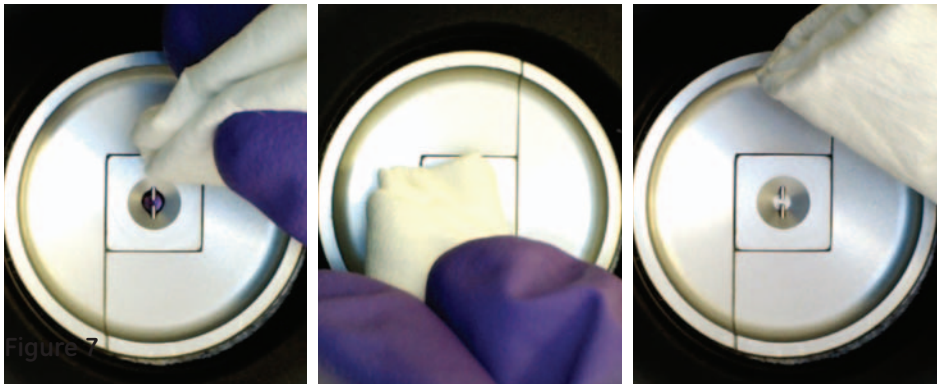


Figure 7: How to clean the sample port

Step 5

Pipette sample into port exactly as performed with the reference as shown in figure 4a, 4b and 6.

3.1. Cleaning and general maintenance

Before cleaning the case of the instrument, switch off the instrument and disconnect the power cord.

Clean all external surfaces using a soft damp cloth. A mild liquid detergent may be used to remove stubborn marks.

The sample port can be cleaned as follows:

- It is recommended that, before starting, the sample port is wiped with a lint free tissue dampened with distilled water.
- For routine use, wiping the sample port with a dry, lint-free tissue between samples, is sufficient to remove sample.
- Periodic cleaning of the sample port is recommended after around 100 samples, or if using sticky protein samples. In this case, a quick wash with a laboratory cleaning detergent is sufficient to remove build up from proteins and DNA. Remove detergent by pipetting on distilled water (maximum volume 25 µl) and removing with a lint-free tissue.
- **Use of spray or squeezy bottles should be avoided, as these flood the sample area.**

3.2. Return for repair

The responsibility for decontamination of the instrument lies with the customer. The case may be cleaned with mild detergent or an alcohol such as ethanol or Isopropanol.

Examination or repair of returned instruments cannot be undertaken unless they are accompanied by a decontamination certificate signed by a responsible person.

This could be of the following form:-

I certify that this instrument and any accessories have been decontaminated to remove any biological, chemical or radioactive hazards and are now safe for handling and transportation.

I certify that no biological, chemical or radioactive hazards are presented by this instrument and accessories due to past use or environment.

Signed: .....

Name (printed): .....

Position of Authority: .....

Date ...../...../.....

On behalf of

3.3. Lamp replacement

The Xenon lamp should not need replacement until used for several years. In the unlikely event that it fails, this should be undertaken by a service engineer from your supplier.

3.4. Replacing the printer paper

Paper for the printer (20 rolls) is available from your supplier under part number 28918226.

Step 1 Keep the power on. Lift off the printer cover.

Step 2 Lock the platen (horizontal position) and feed the paper into the slot. The drive will engage automatically and take up the paper.

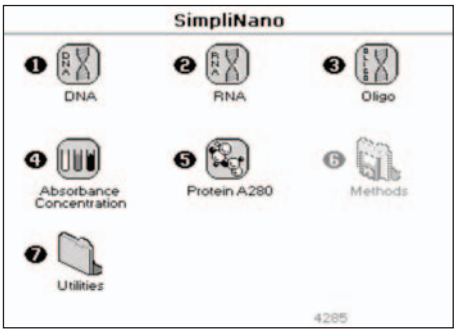
Step 3 It may help to release the platen lock (turn flat green catch clockwise) and turn the green knob manually.

Step 4 Replace the cover.

4. APPLICATIONS

SUMMARY:

Function	Keypad Number	Description
1 DNA	1	Concentration and purity check for DNA samples.
2 RNA	2	Concentration and purity check for RNA samples.
3 Oligo	3	Concentration and purity check for Oligonucleotide samples.
4 Absorbance / Concentration	4	Concentration measurement at a single wavelength based on a factor entered or calculated from a single standard.
5 Protein A280	5	Concentration and purity check for protein samples.
6 Methods	6	Saved methods.
7 Utilities	7	Instrument set up options.



4.1 Nucleic Acid Theory

- Nucleic acids can be quantified at 260 nm because at this wavelength there is a clearly defined maximum peak. A 50 µg/ml DNA solution, a 40 µg/ml RNA solution and 33 µg/ml solution of a typical synthetic Oligonucleotide all have an optical density of 1.0 A in a 10 mm pathlength cell. These factors (50, 40 and 33 respectively) can be inserted into the formula (1) below, although they do vary with base composition and this can be calculated more precisely if the base sequence is known.
  (1) Concentration = Abs260 * Factor (where * means “multiplied by”)
- SimpliNano will default to factors 50 for double stranded DNA, 40 for RNA and 33 for single stranded DNA and Oligonucleotides. It also allows manual compensation for dilution - aided by a dilution calculator.
- Nucleic acids extracted from cells are accompanied by protein and extensive purification is required to remove the protein impurity. The 260/280 nm Absorbance ratio gives an indication of purity, however it is only an indication and not a definitive assessment. Pure DNA and RNA preparations have expected ratios of ≥ 1.8 and ≥ 2.0 respectively. Deviations from this indicate the presence of impurity in the sample, but care must be taken in the interpretation of results.
- The 260 nm reading is taken near the top of a broad peak in the Absorbance spectrum for nucleic acids, whereas the 280 nm reading naturally occurs on a steep slope, where small changes in wavelength will result in large changes in Absorbance. Consequently, small variations in wavelength accuracy have a much larger effect at 280 nm than at 260 nm. It follows therefore that the 260/280 ratio is susceptible to this effect and users are warned that spectrophotometers of different designs may give slightly different ratios.
- In practice, concentration also affects the 260/280 ratio as the individual readings approach the instrument’s detection limit. If a solution is too dilute, the

280 nm reading shows a greater proportional interference from background and as the divisor becomes smaller, has a disproportionate effect on the final result.

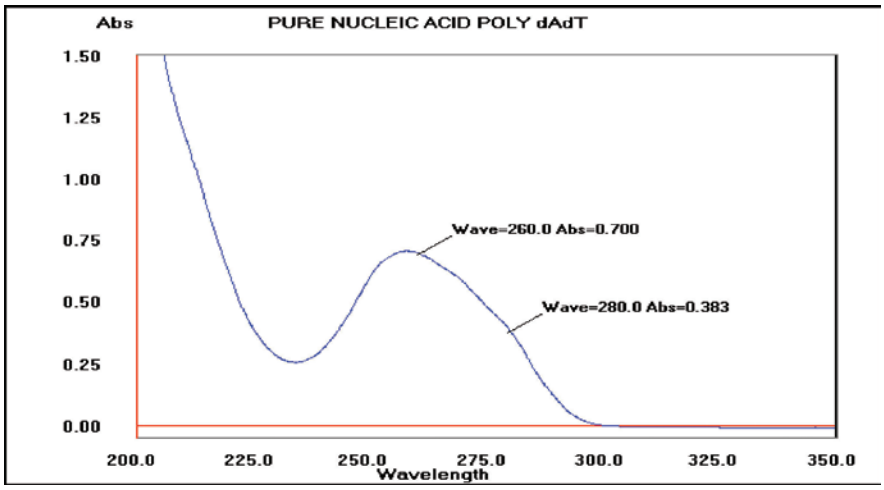
It is advisable to ensure that the Abs260 value is greater than 0.1 A for accurate measurements.

- An elevated Absorbance at 230 nm can also indicate the presence of impurities. 230 nm is near the Absorbance maximum of peptide bonds and may also indicate interference from common buffers such as Tris and EDTA. When measuring RNA samples, the 260/230 ratio should be > 2.0. A ratio lower than this is generally indicative of contamination with guanidinium thiocyanate, a reagent commonly used in RNA purification, which absorbs over the 230–260 nm range. A wavelength scan of the nucleic acid is particularly useful for RNA samples.
- SimpliNano displays the individual Absorbance values and the 260/280 and 260/230 ratios on the left side of the screen and attention should be given to these as well as the final result on the right hand side.

Use of Background Correction

- Background Correction at a wavelength well away from the nucleic acid or protein peaks is often used to compensate for the effects of background absorbance. The procedure can adjust for the effects of turbidity, stray particulates and high-Absorbance buffer solutions.
- SimpliNano uses background correction at 320 nm by default for nucleic acid measurements. It is particularly recommended since very small samples are susceptible to stray particulates. The background function toggles On and Off with either left/right arrows from the relevant page.
- When background correction is on, the results will be different from when it is off as Abs320 is subtracted from Abs260 as shown in the equations below:

Concentration	=	(Abs260 – Abs320) * Factor
Abs ratio 260/280	=	(Abs260 – Abs320) / (Abs280 –Abs320)
Abs ratio 260/230	=	(Abs260– Abs320) / (Abs230 –Abs320)



Typical spectral scan of a Nucleic Acid:-

Note:

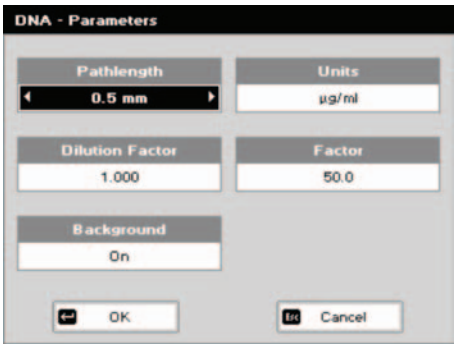
- The Absorbance maximum near 260 nm and Absorbance minimum near 230 nm.
- The flat peak near 260 nm and steep slope at 280 nm.
- There is very little Absorbance at 320 nm.

Sample Type	Lower limit of Detection	Upper Limit of Detection	Reproducibility (Based on 10 replicate measurements)
dsDNA	4.0 ng/µl	2500 ng/µl	<100 ng/ul: Tolerance of +/- 2 ng/ul 100 ng- 2500 ng /µl: Tolerance of +/- 2%

Table 1: Limits of Detection for DNA

4.2. DNA measurement

The procedure is as follows:



Step 1

Press ***1*** to select ***DNA-Parameters*** mode.

Step 2 (dilution factor known)

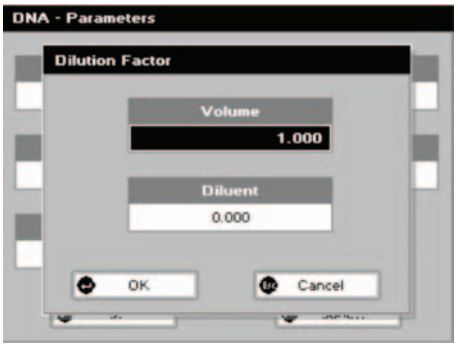
Enter the dilution factor using the keypad numbers. (range 1.00 to 9999).

Use the ***C*** button to backspace and clear the last digit entered.

OR

Step 2 (calculate dilution factor)

Press to enter the ***Dilution Factor*** screen (see second parameter screen to the right).



Enter the volume of the sample using the keypad numbers (range 1.00 to 9999).

Press the ***down*** arrow.

Enter the volume of the diluent using the keypad numbers (range 1.00 to 9999).

Press ***OK*** to calculate the dilution factor and return to the ***DNA-Parameters*** screen.

OR press ***Cancel*** to cancel the selections and return to the ***DNA-Parameters*** screen.

Step 3

Background Correction at 320 nm will be as default. It may be turned off with the ***left*** or ***right*** arrows.

Press the ***down*** arrow.

Step 4

Select the units of measurement using the ***left*** and ***right*** arrows.
Options: µg/ml, ng/µl, µg/µl.

Press the ***down*** arrow.

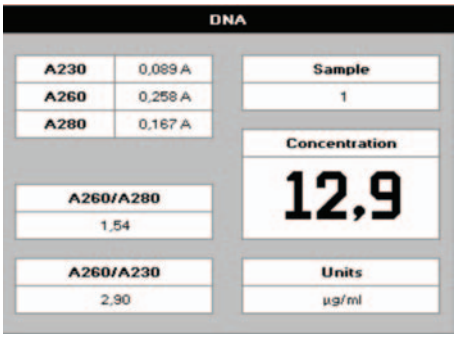
Step 5

Enter the factor using the keypad numbers. Default value is 50, the range is 0.01 to 9999.

Step 6

Press ***OK*** to enter the ***Results*** screen and begin taking measurements.
OR Esc to return to the ***Home*** screen.

Results Screen



Step 7

Pipette the reference sample into the sample port. Press Key. This reference will be used for all subsequent samples until changed. If ***QA*** is switched on the reference sample will need to be replaced and the key pressed again.

Step 8

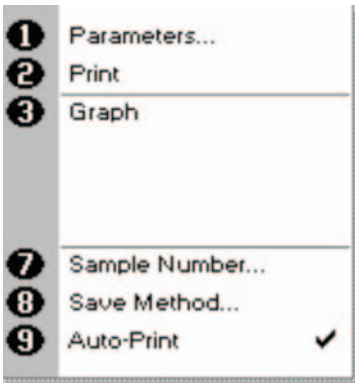
Clean the sample port, pipette the sample into the port. Press . This measures at the selected wavelengths and displays the results.

Repeat step 8 for all samples.

Press Esc to return to the ***Home*** page.

OR press to display the following options.

Options (select using keypad numbers)



❶ Return to ***DNA-Parameters*** screen (step 1 above).

❷ Print result via selected method.

❸ Toggle Graph on/off. The graph shows a wavescan plot across the range 220 nm to 320 nm with cursors denoting 230, 260, 280 and (if Background Correction selected) 320 nm.

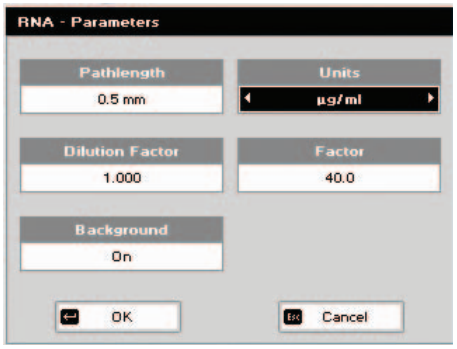
❼ ***Sample Number*** – add a prefix to the sample number and reset the incrementing number to the desired value.

❽ ***Save Method*** – use the alpha-numeric keys to enter a name for the method and press ***Save*** ↵.

❾ ***Auto-Print*** – toggles Auto-Print on/off.

Exit Options by pressing Esc or wait.

4.3. RNA measurement



The procedure is as follows:

Step 1

Press ***2*** to select ***RNA-Parameters*** mode.

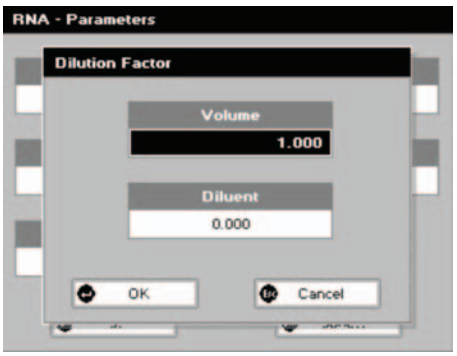
Step 2 (dilution factor known)

Enter the dilution factor using the keypad numbers. Range 1.00 to 9999. Use the ***C*** button to backspace and clear the last digit entered.

OR

Step 2 (calculate dilution factor)

Press ☰ to enter the ***Dilution Factor*** screen (see second image to the right).



Enter the volume of the sample using the keypad numbers. (range 0.01 to 9999).

Press the ***down*** arrow.

Enter the volume of the diluent using the keypad numbers (range 0.01 to 9999).

Press ***OK*** ↵ to calculate the dilution factor and return to the ***Parameters*** screen.

OR press Esc to cancel the selections and return to the ***Parameters*** screen.

Step 3

Select whether the background correction at 320 nm is used or not with the ***left*** and ***right*** arrows.

Press the ***down*** arrow.

Step 4

Select the ***Units*** of measurement using the ***left*** and ***right*** arrows.
Options: µg/ml, ng/µl, µg/µl.

Press the ***down*** arrow.

Step 5

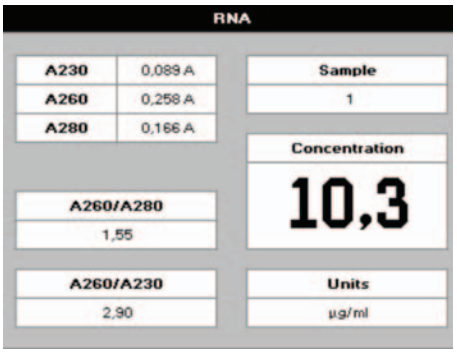
Enter the factor using the keypad numbers. The default value is 40, the range is 0.01 to 9999.

Step 6

Press ***OK*** ↵ to enter the ***Results*** screen and start taking measurements.

OR Esc to return to the ***Home*** page.

Results Screen



Step 7

Pipette the reference sample. Press Key. This reference will be used for all subsequent samples until changed. If ***QA*** is switched on the sample will need to be replaced and the key pressed again.

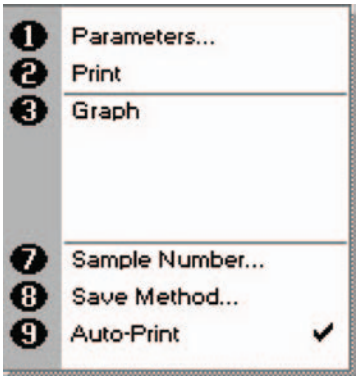
Step 8

Clean the sample port and pipette sample. Press to measure at the selected wavelengths and display results.

Repeat step 8 for all samples.

Press Esc to return to the ***Home*** page.

OR press to display the following options.



Options (select using keypad numbers)

❶ Return to ***Parameters*** screen (step 1 above).

❷ Print result via selected method.

❸ Toggle Graph on/off. The graph shows a wavescan plot across the range 220 nm to 320 nm with cursors denoting 230, 260, 280 and (if Background Correction selected) 320 nm.

❼ ***Sample Number*** – add a prefix to the sample number and reset the incrementing number to the desired value.

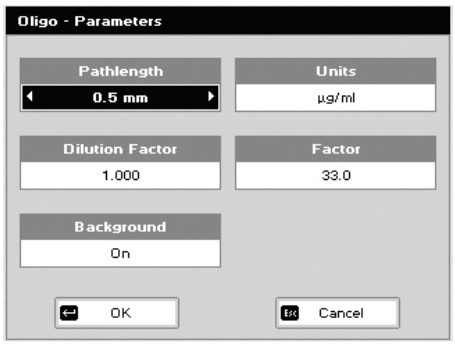
❽ ***Save Method*** – use the alpha-numeric keys to enter a name for the method and press ***Save***.

❾ ***Auto-Print*** – toggles Auto-Print on/off.

Exit Options by pressing Esc, or wait.

4.4. Oligo

The procedure is as follows:



Step 1

Press ***3*** to select ***Oligo*** mode.

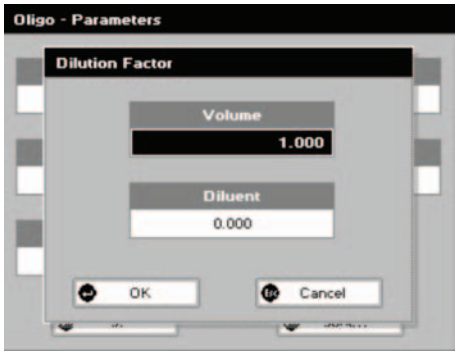
Step 2 (dilution factor known)

Enter the dilution factor using the keypad numbers (range 1.00 to 9999). Use the ***C*** button to backspace and clear the last digit entered.

OR

Step 2 (calculate Dilution Factor)

Press to enter the ***Dilution Factor*** screen.



Enter the volume of the sample using the keypad numbers (range 1.00 to 9999).

Press the ***down*** arrow.

Enter the volume of the diluent using the keypad numbers (range 1.00 to 9999).

Press ***OK*** to calculate the dilution factor and return to the ***Parameters*** screen.

OR press Esc to cancel the selections and return to the ***Parameters*** screen.

Step 3

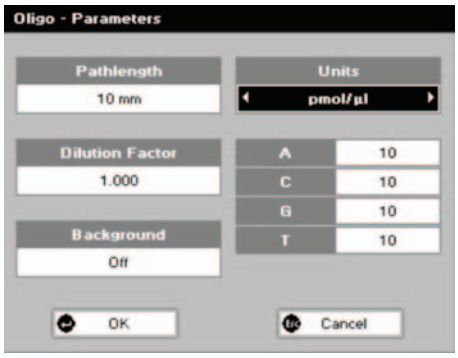
Select whether the background correction at 320 nm is used or not with the ***left*** and ***right*** arrows.

Press the ***down*** arrow.

Step 4

Select the units of measurement using the ***left*** and ***right*** arrows. Options: µg/ml, ng/µl, µg/µl and pmol/µl. If pmol/µl is selected the factor changes to a selection table denoting the ratios of the 4 bases in the structure.

Press the ***down*** arrow.



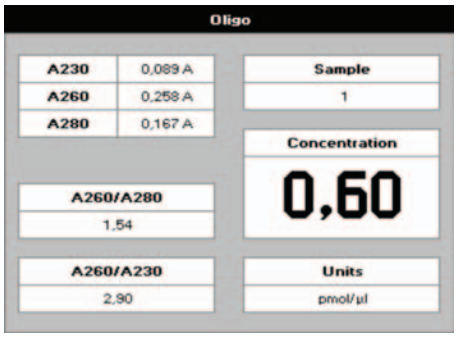
Step 5 (units not pmol/µl)

Enter the factor using the keypad numbers. Default value is 33, range is 0.01 to 9999.

OR

Step 5 (units pmol/µl)

Enter the proportions of bases present using the keypad numbers and ***up*** and ***down*** arrows to move between boxes. Default is 10 for each, range is 0 to 9999.



Step 6

Press ***OK*** to enter the ***Results*** screen and start making measurements.

OR

Esc to return to the ***Home*** page.

Results Screen

Step 7

Pipette the reference sample into the sample port. Press Key. This reference will be used for all subsequent samples until changed. If ***QA*** is switched on the reference sample will need to be replaced and the key pressed again.

Step 8

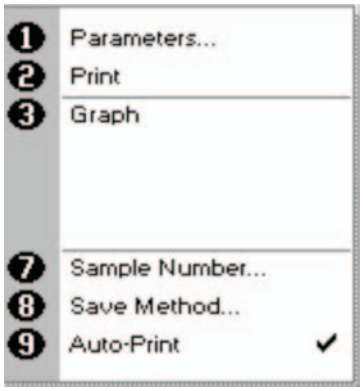
Clean the sample port, pipette the sample and press . This measures at the selected wavelengths and displays the results. The ratio of wavelengths 1 and 2 absorbencies are calculated (both corrected by the background wavelength value if selected). Gives concentration based on absorbance at wavelength 1.

Repeat step 8 for all samples.

Press Esc to return to the ***Home*** page.

Press to display the following options.

Options (select using keypad numbers)



❶ Return to ***Parameters*** screen (step 1 above).

❷ Print result via selected method.

❸ Toggle Graph on/off. The graph shows a wavescan plot across the range 220 nm to 320 nm with cursors denoting 230, 260, 280 and (if Background Correction is selected) 320 nm.

❼ ***Sample Number –*** add a prefix to the sample number and reset the incrementing number to the desired value.

❽ ***Save Method –*** use the alpha-numeric keys to enter a name for the method and press ***Save*** .

❾ ***Auto-Print –*** toggles Auto-Print on/off.

Exit options by pressing Esc, or wait.

4.5. Absorbance/Concentration

The procedure is as follows:

Step 1
Press **4** to select ***Absorbance/Concentration*** mode.

Step 2
Select the wavelength using either arrow or alphanumeric keys.

Press the ***down*** arrow.

Step 3
Enter the measurement mode ***Absorbance/Concentration*** using factor or single standard. If ***Absorbance*** selected press ***OK*** ↵ to enter the ***Results*** screen.
If ***Factor*** or ***Standard*** selected press the ***down*** arrow.

Step 4 (if factor selected)
Enter relevant factor and units. Press ***OK*** ↵ to enter the ***Results*** screen.

Step 5 (If Standard selected)
Enter the concentration and units of the standard to be measured using the keypad numbers. Press ***OK*** ↵ to enter the ***Results*** screen.

Step 6
Pipette the reference sample. Press ⊣ key. This reference will be used for all subsequent samples until changed. If ***QA*** is switched on, the sample will need to be replaced and the ⊣ key pressed again.

Step 7
Pipette the sample and press ⊣. This measures at the selected wavelength and displays the results. Repeat step 7 for all samples.

Step 8 (if Standard selected)
Insert standard, and press ⊣. Change the concentration value of the standard if necessary and press ***OK*** ↵ to measure the standard. Insert sample and press ⊣.

This measures at the selected wavelength and displays the results. Repeat for all samples.

Press ☰ to display the following options.

Press ***Cancel*** Esc to cancel selections and return to the ***Home*** page.

Options (select using key pad numbers).

❶ Return to ***Parameters*** screen (step 1 above).

❷ Print result via selected method.

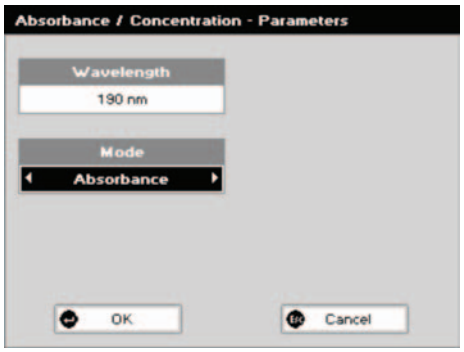
❸ Run Standard.

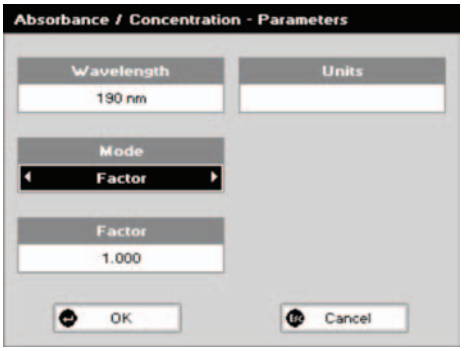
❼ ***Sample number –*** add a prefix to the sample number and reset the incrementing number to the desired value.

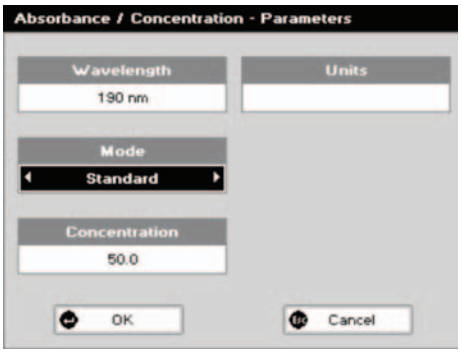
❽ ***Save method –*** use the alpha-numeric keys to enter a name for the method and press ***Save*** ↵.

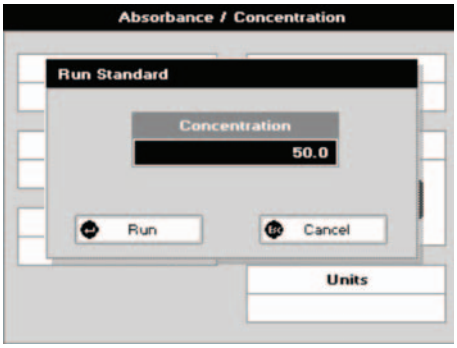
❾ ***Auto-print –*** toggles auto-print on/off.

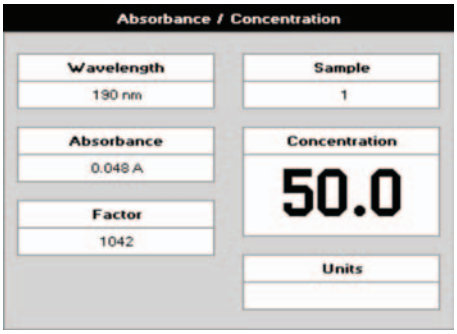
Exit options by pressing Esc or wait.

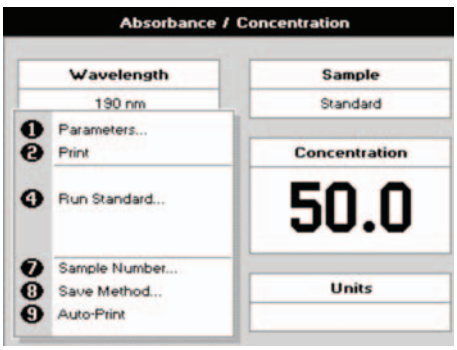












4.6. Protein determination at 280 nm

Sample Type	Lower limit of Detection	Upper Limit of Detection	Reproducibility (Based on 10 replicate measurements)
BSA	0.12 mg/ml	50 mg/ml	<1 mg/ml: Tolerance of +/- 0.06 mg/ml 1 mg/ml- 5 -mg/ml: Tolerance of +/- 2%

Table 2: Limits of Detection for BSA

Proteins can be determined in the near UV at 280 nm due to absorption by tyrosine, tryptophan and phenylalanine amino acids. The Abs280 varies greatly for different proteins due to their amino acid content and consequently the specific Absorption value for a particular protein must be determined.

- The presence of nucleic acid in the protein solution can have a significant effect due to strong nucleotide Absorbance at 280 nm.
- The Protein A280 application, has three modes, which can be selected depending on the extinction coefficient of the reference protein being used.

  A) For BSA the extinction coefficient is 6.7 AU (E1%) at Abs 280 for a 1% ww solution

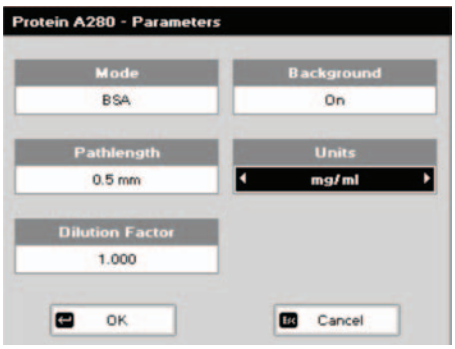
  B) For IgG the extinction coefficient is 13.7 AU (E1%) at Abs 280 for a 1% ww solution

  C) For lysozyme the extinction coefficient is 26.4 AU (E1%) at Abs 280 for a 1% ww solution.
- Rapid measurements such as this at Abs280 are particularly useful after the isolation of proteins and peptides from mixtures using spin and HiTrap columns by centrifuge and gravity, respectively.

Use of Background Correction

- Background Correction at a wavelength well away from the protein peak is used to compensate for the effects of background Absorbance. The procedure can adjust for the effects of turbidity, stray particulates and high-Absorbance buffer solutions.
- SimpliNano uses background correction at 340 nm by default protein measurements. It is particularly recommended since very small samples are susceptible to stray particulates. The Background function toggles On and Off with either left/right arrows from the relevant page.
- When background correction is on, the results will be different from when it is off as Abs340 is subtracted from Abs280 prior to reporting.

4.7. Protein A280



Step 1

Press ***5*** to select ***Protein A280***.

Step 2

Select the ***Mode***. Options are, BSA, IgG, Lysozyme.

Press the ***down*** arrow.

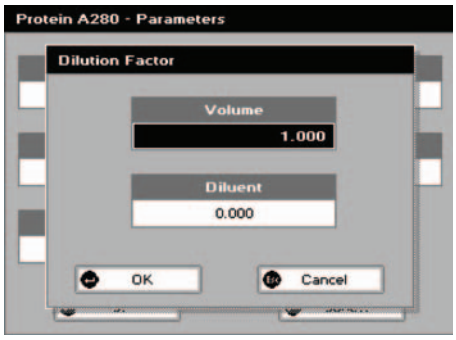
Step 3 (dilution factor known)

Enter the dilution factor using the keypad numbers (range 1.000 to 9999).
Use the ***C*** button to backspace and clear the last digit entered.

OR

Step 4 (calculate dilution factor)

Press to enter the dilution factor screen, shown to the right.



Enter the volume of the sample using the keypad numbers (range 0.001 to 9999).

Press the ***down*** arrow.

Enter the volume of the diluent using the keypad numbers (range 0.001 to 9999).

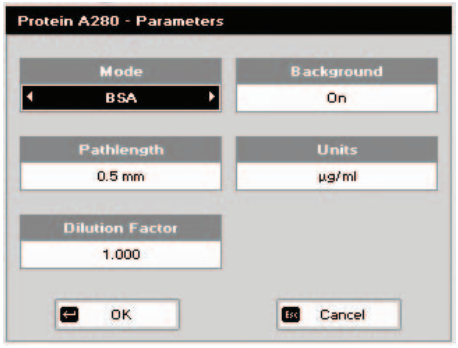
Press ***OK*** to calculate the dilution factor and return to the ***Parameters*** screen.

OR press Esc to cancel the selections and return to the ***Parameters*** screen.

Step 5

Select whether Background Correction is to be used or not with the ***left*** and ***right*** arrows.

Press the ***down*** arrow.



Step 6

Select the Units of measurement using the ***left*** and ***right*** arrows.
Options: mg/ml, µg/ml, ng/µl and µg/µl.

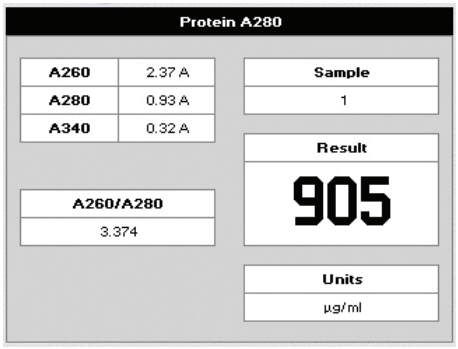
Step 7

Press ***OK*** to enter the ***Results*** screen.

OR

Cancel Esc to return to the ***Protein*** screen.

Results Screen



Step 8

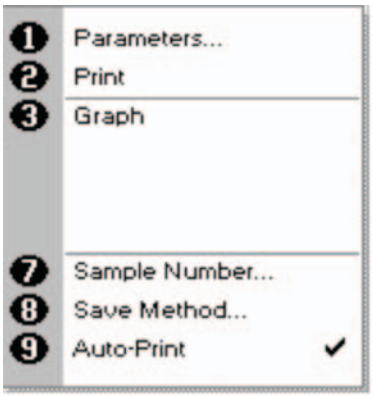
Pipette on the reference sample and press the key. This reference will be used for all subsequent samples until changed. If ***QA*** is switched on, the sample will need to be replaced and the key pressed again.

Step 9

Clean the sample port, pipette the sample and press . This measures at the selected wavelength and displays the results.

Repeat step 9 for all samples.

Press to display the following options.

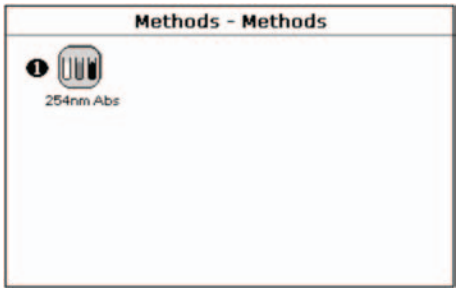


Options (select using keypad numbers)

1. Return to ***Parameters*** screen (step 1 above).
2. Print result via selected method.
3. Toggle Graph on/off. The graph shows a wavescan plot across the range 250 nmto 350 nm.
7. ***Sample Number –*** add a prefix to the sample number and reset the incrementing number to the desired value.
8. ***Save Method –*** use the alpha-numeric keys to enter a name for the method and press ***Save***.
9. ***Auto-Print –*** toggles Auto-Print on/off.

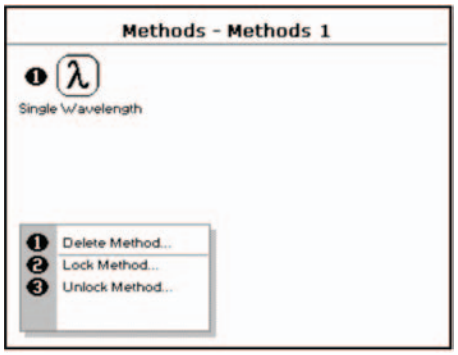
Exit Options by pressing Esc ,or wait.

5. METHODS FOLDER



Press ***6*** to enter the ***Methods*** folder. This folder is the storage locations for any user modified applications **(Methods)** that are saved using the ***Options*** menu and is accessible from the ***Home*** page.

Up to 9 ***Methods*** may be stored, saved methods can be locked, unlocked and deleted using the ***Options*** menu. Select the method by pressing the relevant key pad number and then press the key.



Delete Method

Press ***1*** to select ***Delete Method***.

Select the method to be deleted using the left and right arrows.

Press ↵ to delete the method OR ***Cancel*** Esc to return to ***Favourites/Methods*** folder.

Lock Method

Press ***2*** to select ***Lock Method***.

Select the method to be locked using the ***left*** and ***right*** arrows.

Press the ***down*** arrow.

Select a pass code using the keypad numbers or ***left*** and ***right*** arrows.

Press ↵ to lock the method OR ***Cancel*** Esc to return to the ***Methods*** folder.

Unlock Method

Press ***3*** to select ***Unlock Method***.

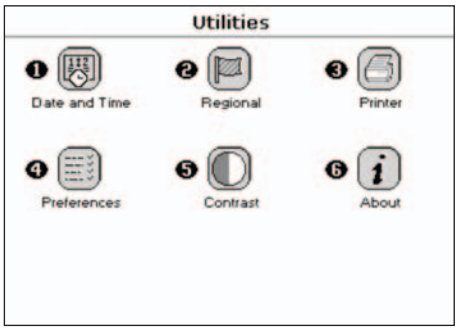
Select the method to be unlocked using the ***left*** and ***right*** arrows.

Press the ***down*** arrow.

Enter the pass code using the keypad numbers or ***left*** and ***right*** arrows.

Press ↵ to unlock the method OR ***Cancel*** Esc to return to the ***Methods*** folder.

6. UTILITIES

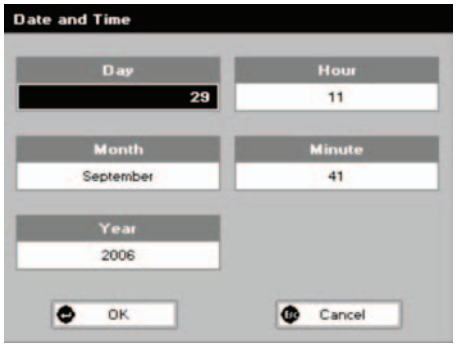


Press ***7*** to enter the ***Utilities*** folder.

Summary

Function	Keypad number	Description
1 Date and Time	1	Set correct date and time
2 Regional	2	Select preferred language and number format
3 Printer	3	Printer/output options
4 Preferences	4	Select screen layout (themes) and history
5 Contrast	5	Adjust screen contrast & brightness
6 About	6	Serial number and software version

6.1. Date and Time



Enter the day using the keypad numbers or left and right arrows.

Press the ***down*** arrow.

Enter the Month.

Press the ***down*** arrow.

Enter the Year.

Press the ***down*** arrow.

Enter the Hour.

Press the ***down*** arrow.

Enter the Minute. Seconds are zeroed when ***OK*** is pressed.

Press ***OK*** ↵ to store the settings and return to the ***Utilities*** screen.

OR

Press ***Cancel*** Esc to return to the ***Utilities*** screen without storing the time.

6.2. Regional

Sets Language and Number Format.

The procedure is as follows:

Select a Language. Options are English, French, German, Spanish, Italian, Chinese or Japanese.

Press the ***down*** arrow.

Set the decimal point style. Options are "," or ".".

Press ***OK*** ↵ to store the settings and return to the ***Utilities*** screen

OR

Press ***Cancel*** Esc to return to the ***Utilities*** screen without storing the settings.

6.3. Printer

Sets up printing options.

The procedure is as follows:

Select whether ***Auto-Print*** is on or off using the left and right arrows. When ***Auto-Print*** is on, the results are automatically printed after a measurement is taken. When it is off, printing has to be initiated manually. This can also be set using the Options key in each application or method. The default is Off.

Press the ***down*** arrow.

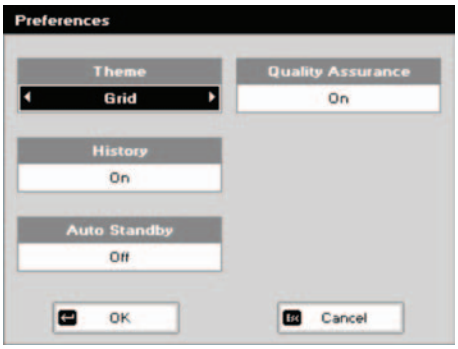
Select how the data are sent. Options are Built in (internal printer), or to a computer via USB port.

Press ***OK*** to store the settings and return to the ***Utilities*** screen.

OR

Press ***Cancel*** Esc to return to the ***Utilities*** screen without storing the settings.

6.4. Preferences



Sets user Preferences.

Define the screen layout of folders. Options are either a grid format (default) or a list.

Press the ***down*** arrow.

Select whether to use previously entered parameters when the instrument is switched on or to use defaults.

Press the ***down*** arrow.

Select whether to use a standby mode after defined periods. Options are 1 hour, 2 hours, at night or off.

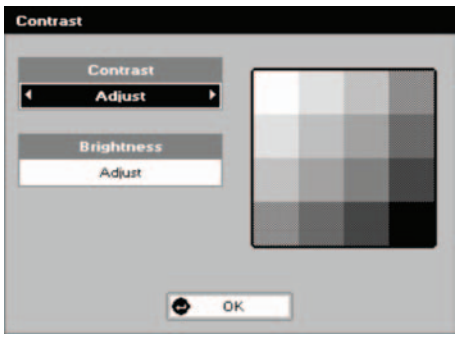
Select whether you want ***Quality Assurance*** mode on/off. ***QA*** mode prompts you to run a 2nd Reference and checks that both references agree within a specified tolerance.

Press ***OK*** to store the settings and return to the ***Utilities*** screen**.**

OR

Press ***Cancel*** Esc to return to the ***Utilities*** screen without storing the settings.

6.5. Contrast



Ambient temperature can affect the display. This function can optimise the display for local conditions.

The procedure is as follows:

Adjust the contrast using the ***left*** and ***right*** arrows to scroll though the levels.

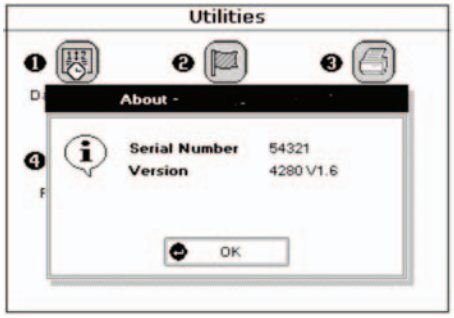
Press the ***down*** arrow. briefly to move down.

Adjust the brightness similarly.

Press the ***down*** arrow.

Press ***OK*** to store the settings and return to the ***Utilities*** screen.

6.6. About



Displays the instrument serial number and software version.

Press ***OK*** to close the window and return to the ***Utilities*** screen.

7. ACCESSORIES INSTALLATION

7.1. Printer installation

The user can install a printer.

Step 1.
Remove the power cable and turn the instrument over onto a soft surface, taking care not to damage the sampling head.

Release the outermost screws using the Allen key provided.

Step 2.
Turn the instrument back over and remove the accessory covers.

Step 3.
Attach the printer cable.

Step 4.
Lower the printer onto the locating bosses.

Step 5.
Replace the top cover plate, invert the instrument and replace the cap head screws at locations A and B shown in step 1.

Step 6.
Switch the instrument on and go to utilities/instrument/preferences and select the Built-in printer.

7.2. After sales support

For all technical support and advice on SimpliNano please contact your supplier.

GE Healthcare's website is: http://www.gelifesciences.com
or http://www.gelifesciences.com/contact

Support agreements that help you to fulfil the demands of regulatory guidelines concerning GLP/GMP are available.

- Calibration, certification
- Certificated engineers and calibrated test equipment
- Approved to ISO 9001 standard

Choice of agreement apart from break down coverage can include;

- Preventative maintenance
- Certification

Accessories

Built-in Printer accessory	28-9182-27
Spare paper for printer (20 rolls)	28-9182-26

8. TROUBLESHOOTING

Problem	Possible cause
Negative absorbance readings	Sample measurements will be negative absorbance reading if the absorbance value of the reference is higher than the sample. Negative readings can also result if reference and sample are interchanged or if the sample is very dilute and close to the absorbance of the reference. Contact your supplier for advice on the minimum concentrations that can be measured.
Unexpected results	• Bubbles or contamination in the sample or reference can result in considerable errors. • Use of incompatible buffers (UV Absorbing) as a reference. • Ensure background correction is switched on. • Trying to measure sample volumes/concentrations outside of the instrument working ranges.
Absorbance higher than expected	• Incorrect sample reference. • Contamination in sample. • Check size and position of droplet. • Ensure background correction is switched on • Possible incorrect optical alignment. Contact technical support.
Absorbance lower than expected	• Incorrect sample reference. • Check sample and reference for contamination. • Check sample and reference samples are not the same. • Check size and position of droplet. • Possible stray light issue. Contact technical support.
Poor reproducibility	• Insufficient sample volume – check pipette calibration. • Concentration of sample too low or too high. • Particulates in sample. Absorbance measurements will not be accurate in turbid samples. • Possible noise or measurement stability issue. Contact technical support.
Instrument start up reported failure	• Check all sample paths are clear and clean – dried on DNA/Protein sample on the Micro-volume head may cause start up calibration errors. • Check original 18 V dc supply is connected and is fully engaged. • Report persistent failures to technical support.
Instrument switches off after calibration	• You may be keeping you finger on the ON/OFF button too long, so that the instrument receives both ON and OFF signals and switches off after the calibration. • Try adjusting the timing of your finger press at switch on. • If the problem persists please contact your supplier.
Absorbance readings stable but different than expected	Remember that the Absorbance displayed is being normalized to a path length of 10 mm. With a 0.5 mm path length the ideal measurement range becomes equivalent when normalised to 2 A to 50 A. For unresolved Absorbance issues contact technical support.
No image on screen	If the lamp can be heard buzzing at startup but there is no image on the screen then the display may be faulty. Please contact your supplier.

8.1. Frequently asked questions

Q. What is the Pathlength of SimpliNano?
A. 0.5 mm.

Q. How accurate is SimpliNano?
A. Typically within 2%.

Q. Does SimpliNano produce results for a continuous spectrum or just selected wavelengths?
A. Within The DNA, RNA, Oligo and protein A280 methods a continuous spectrum is produced.

Q. Do nucleic acids require purification before being measured?
A. Yes. Absorbance measurements are not specific for a particular nucleic acid and will be affected by the presence of other molecules which absorb at 260 nm.

Q. How should the sample port be cleaned?
A. The sample port is easy to clean. Simply wipe with a lint free tissue or cloth. If a difficult or sticky sample or dye was used, the port can be cleaned with a dilute (2%) detergent solution followed by water or isopropanol. See Section 3.1 Cleaning and General Maintenance, Step 4 for details.

Q. Is a simple wipe with a lint free tissue or cloth enough to prevent carryover?
A. Yes. A sample carryover study using 25 mg/ml BSA with successive alternative additions of water and BSA, was repeated 50 times. The data showed no significant change in absorbance readings throughout the study for the water samples or the BSA samples, indicating that there was no carryover. See SimpliNano Datafile 29-1028-98 for details.

For more difficult, sticky, or strongly coloured samples, the port can be cleaned with a dilute (2%) detergent solution, followed by water or isopropanol.

Q. Are there any solvent restrictions?
A. Most common laboratory solvents, including dilute acids, may be used but they should be immediately wiped off the sample head after measuring. The SimpliNano has been tested with methanol, isopropanol (2-propanol), and acetone. However, these solvents may affect the accuracy of the measurement due to their volatility & are therefore not recommended.

Q. Why does the reference (blank) require 2 samples?
A. To set the reference, the software will ask for 2 separate samples to ensure the quality of the reference baseline instrument.

Q. What kind of light source does the SimpliNano use?
A. A pulsed Xenon flash lamp.

Q. How often will the lamp require changing?
A. The lamp should last for several years of use & comes with a 3 year warranty. If it does need changing, this should be done by a service engineer from your supplier.

Q. Is the flash lamp on continuously, or only when performing a measurement?
A. The lamp is only on when taking measurements.

Q. Does SimpliNano require a computer to operate?
A. No, SimpliNano operates as a stand alone instrument and data can be transferred via USB stick. If operation via a computer is critical it can be connected to a PC and operated using either PVC or Datrys software.

Q. Can I get data exported in Excel format?
A. Using the PVC software application, data may be stored either manually or automatically in a variety of data formats which include : as an Excel spreadsheet (Excel must be installed on the PC), Extensible Meta Format (an EMF) graphics file, a comma delimited (csv) data file, a tab delimited (txt) file, Rich Text Format (RTF) or in native PVC format.

Q. What volume of sample can I use with the SimpliNano?
A. Volumes as low as 1 µl can be used. A sample volume of 2 µl is recommended to minimize pipetting errors affecting results.

Q. What calibration does the SimpliNano require?
A. The SimpliNano has no moving parts so calibration is not required.

Q. How reproducible is the Absorbance data?
A. Specifications for precision and accuracy are below.

Absorbance Precision	Between 0 & 1 A; < 0.005 A or 1% of measurement (whichever is greater)
	1 to 2 A – 2% of reading; above 2 A - 5% of reading
Absorbance Accuracy	± 1% at 257 nm, Potassium Dichromate solution, 0.7 – 0.8 A.

Q. What are the detection limits for SimpliNano.
A. For dsDNA the range is 4.0 ng/µl 2500 ng/µl and for BSA 0.12 mg/ml 50 mg/ml .

Q. What sort of reproducibility should I expect with the SimpliNano?
A. Typically ± 2 ng/µl for dsDNA samples < 100 ng/µl- 2500 ng/ul and ± 2% for samples > 100 ng/µl. For BSA, typically +/- 0.1 mg/ml for samples 1-50 mg/ml and for >1 mg/ml tolerance of +/- 2%.

Q. What purity ratio measurements are available on the SimpliNano?
A. The SimpliNano measures 260/280 and A260/A230 ratios for Nucleic Acids and A260/A280 ratios for proteins.

9. SPECIFICATION AND WARRANTY

Specification

Wavelength range	190–1100 nm
Monochromator	Flat Grating
Wavelength calibration	Automatic upon switch on
Spectral bandwidth	5 nm
Wavelength accuracy	±2 nm
Wavelength reproducibility	±1 nm
Light sources	Pulsed Xenon lamp
Detector	1024 element CCD array
Photometric range	- 0.300 to 2.500 A, 0 to 199%T
Photometric linearity	±0.005 Abs or 1% of the reading, whichever is the greater @ 546 nm
Photometric reproducibility	±0.003 Abs (0 to 0.5 Abs), ±0.007 Abs (0.5–1.0 Abs)
Stray light	<0.5% at 220 nm and 340 nm using $NaNO_2$
Zero stability	±0.01 Abs/hour after 20 min warm up @ 340 nm
Noise	0.005 pk to pk 0.002 RMS
Digital output	USB B port for connection to PC
Dimensions	260 × 390 × 130 mm
Weight	<4.5 kg
Power input	18 Vdc from a 90–250 V, 50/60 Hz, Max 30 VA mains power pack

Specifications are measured after the instrument has warmed up at a constant ambient temperature and are typical of a production unit. As part of our policy of continuous development, we reserve the right to alter specifications without notice.

Warranty

- GE guarantees that the product supplied has been thoroughly tested to ensure that it meets its published specification. The warranty included in the conditions of supply is valid for 24 months only if the product has been used according to the instructions supplied. GE can accept no liability for loss or damage, however caused, arising from the faulty or incorrect use of this product.
- The lamp is warranted for 3 years if the product has been used according to the instructions supplied.

10. Legal

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